Background and Objectives: Understanding human milk composition is critical for setting nutrient recommended intakes (RNIs) for both infants and lactating women. However, nationwide human milk composition remains unavailable in China. Methods and Study Design: Through cross-sectional study, human milk samples from 11 provinces in China were collected and their compositions were analyzed. Nutritional and health status of the lactating women and their infants were evaluated through questionnaire, physical examination and biochemical indicators. Results: A total of 6,481 breast milk samples including colostrum (1,859), transitional milk (1,235) and mature milk (3,378) were collected. Contents of protein, fat, lactose, total solid and energy of more than 4,500 samples were analyzed using a human milk analyzer. About 2,000 samples were randomly selected for 24 mineral analyses. Free B-vitamins including thiamin, riboflavin, pyridoxal, pyridoxamine, nicotinamide, nicotinic acid, flavin adenine dinucleotide (FAD), biotin and pantothenic acid were analyzed in 1,800 samples. Amino acids (~800) and proteins (alpha-lactalbumin, beta-casein, and lactoferrin) were analyzed. In addition, serum retinol and carotenoids, 25(OH)D, vitamin B-12, folic acid, ferritin and biochemical indicators (n=1,200 to 2,000) were analysed in the lactating women who provided the breast milk. Ongoing work: Fatty acids (C4-C24), fat-soluble vitamins and carotenoids, are on-going analysis. Conclusions: A regional breast milk compositional database is at an advanced stage of development in China with the intention that it be available on-line.

Key Words: human milk, composition, database, lactating women, China

INTRODUCTION
It is well known that human milk is the best food for infants. Exclusively breastfeeding is recommended for infants during the first 6 months of life, with continued breastfeeding until 2 years of age or beyond as well as introducing adequate complementary foods from 6 months of age. Breastfeeding can meet the nutrition needs of infants in the first 6 months of life and contribute to adequate growth and development of infants and young children. In addition, breastfeeding can reduce the risk of gastrointestinal infection, respiratory infections, allergies and autoimmune diseases.

Human milk is considered as the gold standard to estimate nutrient recommendations for infants. The required composition and adequate range of nutrients for infant formulas in the US as well as most of the other countries or International standards are based on the composition of human milk. However, how to mimic human milk remains not fully understood. Other outcomes besides the composition need to be assessed including the optimal nutrition and health status of infants.

Publications on “human milk composition” indexed in PubMed have increased steadily, especially in regard to active components and their function as they have been identified in human milk. However, data differ with population diversity, varied sample collection, storage and analytical methods. For example, sample collection methods are often not standardized by stage of lactation, time of day or timings of and between feeds. Sample storage and treatment also vary among studies of human milk composition in regard to number of freeze-thaw cycles, duration and temperature of storage.

It has been shown that the variability of human milk components is quite wide and many factors can affect them. For example, human-milk composition is affected by stage of lactation, numerous genetic factors (contributing to individual variability), maternal age, weight and diets, diurnality, and environmental factors. Protein content decreases during the postpartum period. Fat content is consistently higher in hind milk and significantly lower in the night and morning feeds (diurnal pattern) compared with the afternoon or evening feeds. Maternal age and region may also be related to some components of breast milk. Overweight mothers might have a lower milk protein concentration. Maternal diet could affect the composition such as lipids and fat-soluble vitamins which might be more susceptible to long-term ma-
ternal nutritional status than short-term dietary fluctuation. For example, maternal protein intake may affect the lipid concentration of breast milk. Micronutrients, including water-soluble vitamins and some trace elements (e.g., selenium, iodine), vary in human milk depending on the maternal dietary intake and body stores. Therefore, understanding these variables is important for estimating the requirements or adequate intakes for some nutrients.

Chinese dietary patterns have changed dramatically in recent times with transitions in quality and quantity. These changes might directly affect some breast milk components and, therefore, potentially affect infant growth and development. However, there have been no nationwide human milk composition data available in China. The present report is about the endeavours in China to analyze the nutrition-related composition of breast milk across the country, to estimate the dietary intakes of lactating women in relation to their nutritional and health status, and to the growth and development of their infants. Finally, we signal the intention to make this information available on-line.

METHODS

Study design

This is a cross-sectional study. Eleven provinces, municipalities or autonomous regions were selected, including Beijing (North), Shanghai (East), Guangzhou (South), Heilongjiang (Northeast), Gansu (Northwest), Yunnan (Southwest), Shandong (coastal area) and Zhejiang (coastal area), Inner Mongolia (Mongolian ethnicity), Xinjiang Uygur (Uygur ethnicity), Guangxi (Zhuang ethnicity). One to four counties were selected from each province, and 340 lactating women and their breastfeeding babies (less than 11 months of age) were recruited from each county except 100 dyads of mother and infant from Inner Mongolia, Xinjiang Uygur and Guangxi.

Study sites had contrasting characteristics. Beijing, Shanghai and Guangzhou Municipalities were considered as large cities where two sites each (urban and peri-urban) were selected to collect information from 680 dyads of lactating women and their breastfeeding infants. Two rural sites and two urban sites (small-medium cities) were selected from Heilongjiang province. Three rural sites were selected from Gansu province to reflect the status of Han, Hui and Tibetan ethnicities and three sites were selected from Yunnan Province to reflect the status of Han, Bai and Dai ethnicities. Weihai city in Shandong Province and Zhoushan city in Zhejiang Province belong to coastal areas. We selected one representative site from each of Xinjiang, Neimeng and Guangxi Autonomous regions to collect milk samples and some information from 100 dyads of lactating women and their babies (one to six months of age) to evaluate the status for the Uygur, Mongolian and Zhuang ethnic groups.

Subjects

This study was approved by the Ethics Committee of the National Institute of Nutrition and Food Safety of Chinese Center for Disease Control and Prevention (CDC). Written informed consent was obtained from all lactating women.

A total of 6,481 dyads of healthy Chinese lactating mothers and their breastfeeding babies within 0-330 days postpartum from 20 sites of 11 Provinces, Municipalities or Autonomous regions in China were recruited for our study.

Inclusion and exclusion criteria

Inclusion criteria were lactating women aged 20-35 years, breastfeeding her baby, self-reported good health, singleton pregnancy, no drinking of alcohol and no smoking, a healthy baby, and signed informed consent. Exclusion criteria included mastitis, infectious disease (specifically tuberculosis, viral hepatitis and HIV infection, but in general as well), cardiovascular disease, metabolic disease (such as diabetes), mental health disorders, ‘cancer’ or other malignant or degenerative diseases, inability to answer questions, currently participation in any study related to nutrition or drug intervention.

Sample size

To ensure that the concentration of nutrients measured in breast milk would provide a range of ±0.05%, the estimated sample size was calculated as follows:

\[
N = \left(\frac{Z_\alpha/2 \times \sigma}{\delta}\right)^2
\]

\(\alpha=0.05\), \(\sigma=\)the standard deviation of each nutrient, \(\delta=0.05\%\).

For example, the required sample size for a large variation of fat content in breast milk was estimated as follows:

Fatty concentration in breast milk was 3.4\% (2.41%-4.21\%): \(n=(1.96 \times 0.5/0.05)^2=385\)

Protein concentration in breast milk was 1.3\% (1.09%-2.19\%): \(n=(1.96 \times 0.45/0.05)^2=312\)

A final sample size of ~ 340 was needed for each survey site, which included 100 colostrum samples and 240 transitional milk and mature milk samples (30 samples × 8 time points). The time points for collected milk were 0-7 days of postpartum (colostrum), and then 8-10 days, 11-13 days, 14-16 days, 17-30 days, 31-90 days, 91-150 days, 151-240 days and 241-330 days. Total sample size for this study was to be around 6,400. (For colostrum, due to small volume milk excreted at one time, more dyads of the lactating women and her baby were recruited).

Field survey

Field work included interview, physical examinations and collection of breast milk. Blood and human milk samples were analyzed in the field and lab.

Interview

During the study visits, trained research staff administered a standardized questionnaire to collect information about lactating women and their breastfeeding babies at local health centers or in temporary assessment clinics set up within the local residential center (village or street committee). The questionnaire included two parts, one for lactating women and the other for her baby. (1) Questionnaire on lactating women: information on socio-economic status, current participation in any study related to nutrition or drug intervention, reported good health, single, smoking or no smoking, drinking of alcohol and no smoking, a healthy baby, and signed informed consent. Exclusion criteria included mastitis, infectious disease such as diabetes, mental health disorders, ‘cancer’ or other malignant or degenerative diseases, inability to answer questions, currently participation in any study related to nutrition or drug intervention.

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ments, and medical history were collected. (2) Questionnaire on breastfeeding infant: birth outcome, breastfeeding status, introduction of complementary foods, other food intake within 24 hours such as infant formula, frequency of complementary food feeding, water and beverage intake, were collected.

Physical examinations
Before or after the interview, weight, height, waist and hip circumferences were measured for all lactating women using standard anthropometric procedures. Height, waist and hip circumferences were measured to the nearest 0.1 cm, and body weight to the nearest 0.1 kg. An anthropometric meter (WS-RT-1B, 50 g of accuracy, Wuhan Computer Software Development Company, China) and a standard measurement tape were used to measure body weight, length, and head circumference of children wearing light clothes, respectively. Before starting field work, weight and height scales were checked with calibrated objects.

Collection and preparation of breast milk samples
One full-breast was emptied using a portable automatic breast pump (HNR/X-2108Z, Shantou, Guangdong, China) in the morning (9 am to 11 am) from November 2011 to June 2013. Milk samples collected at home for some lactating women within one month after delivery were immediately put into the ice box, or 4°C refrigerator. After the completion of the interview in field site, the milk samples were immediately transferred to the local laboratory.

In the field or local laboratory, the human milk samples were gently up-down shaken in the milk bottle for ~10 times, and then fresh milk samples (~3 mL) were taken using a disposable syringe (5 mL) for determining protein, lipids, carbohydrates, energy and total solid using HMA (HMA, Miris, Sweden) in the field site.12,20,21 The remaining milk samples were aliquoted into five of 10 mL centrifuge tubes (each tube had about 9-10 mL), the excess portion was dispensed to a plurality of storage 50 mL tube) for subsequent analysis. Colostrum samples were stored in a 10 mL tube. All samples were wrapped with aluminum foil to avoid sunlight exposure. The aliquoted samples were stored at -20°C refrigerator in the field. After field study was completed, the samples were shipped to our Beijing laboratory under the frozen status and were stored at -80°C freezer until analysis.

Laboratory analysis
A 5 mL fasting blood sample was drawn from an antecubital vein of each lactating women after 30 days of delivery by phlebotomists in the morning. Hemoglobin (Hb) concentration in whole blood from lactating women was measured using the HemoCue (HB 301, HemoCue AB, Angelholm, Sweden) at field sites. Hb concentration of whole blood of each infant after 3 month of age, taken from the left middle finger tip, was also analyzed using the hemocue at field sites. Anemia was defined as Hb level of <120 g/L for lactating women and <110 g/L for infants. At the sites with an altitude over 1,000 meters, the anemia prevalence was corrected using WHO recommended altitude formula.

The blood samples were collected in heparin separation tubes and kept for 45 minutes under the light protection condition. Then blood samples were centrifuged at 3,000 rpm for 10 minutes at room temperature. The plasma fraction was collected and white blood cells and red blood cell components were separated. Then all samples were kept at -20°C~30°C freezer at field sites. Blood glucose concentration of lactating women was measured in field site.

Blood biochemical indicators and nutritional biomarkers
Maternal blood biochemical indicators were measured with an automated biochemical analyzer (Hitachi 7600, Tokyo, Japan) in Beijing Center for Disease Control and Prevention, according to the international quality standard. Reagents from the same batch were used to minimize laboratory variability (n=1,983). These indicators included alanine aminotransferase, aspartate aminotransferase, total protein, albumin, alkaline phosphatase, glucose, calcium, phosphorus, cholesterol, triglycerides, high-density lipoprotein cholesterol, low density lipoprotein cholesterol, apolipoprotein A, apolipoprotein B, lipoprotein (a), potassium (K), sodium (Na), chloride (Cl), iron (Fe), zinc (Zn), magnesium (Mg), copper (Cu) and C-reactive protein.

The analyses of other serum micronutrients were conducted by the National Institute for Nutrition and Food Safety for the Chinese CDC. Serum retinol and carotenoids and tocopherol were assessed using a modified HPLC method (Waters 600E, US);22 serum 25-OH-D, vitamin B12, folic acid and ferritin were determined using commercial radioimmunoassay kits (25-OH-D, Dria Sorin, US; ferritin, Northern Institute of Biotechnology, China). Vitamin B-12 and folic acid were analyzed by using electrochemiluminescence methods. Twenty-four elements in milk were determined with inductively coupled plasma mass spectrometry (ICP-MS), which included Na, Mg, K, calcium (Ca), aluminum (Al), Fe, Zn, manganese (Mn), Cu, phosphorus (P), arsenic (As), selenium (Se), molybdenum (Mo), vanadium (V), cadmium (Cd), chromium (Cr), cobalt (Co), nickel (Ni), gallium (Ga), silver (Ag), strontium (Sr), cesium (Cs), barium (Ba) and lead (Pb).23

Free B-vitamins in human milk were measured using modified UPLC-MS/MS which included thiamin, riboflavin, pyridoxine, pyridoxal, pyridoxamine, nicotinic acid and nicotinamide, flavin adenine dinucleotide, biotin, and pantothentic acid.24

Quality control
The expert group, from National Institute for Nutrition and Food Safety of Chinese Center for Disease Prevention, was responsible for formulating and evaluating the questionnaire, the work program, field survey operation and laboratory manuals, creating the data entry program and data management, training the Project Working Group from the Provinces or Municipalities or Autonomous regions in survey methods, skill training for laboratory operations, arranging the transportation, preservation and analysis of milk samples and blood samples collected in the field sites, organizing the training for data entry,
national data integration, data security, analysis and summary of the national survey data.

Province, municipality or autonomous regional CDCs (Centers for Disease Control) organized the survey team in field sites and trained them and carried out the field survey according to the unified survey arrangements and working manual, and were responsible for data collation, verification, entry and transfer to China CDC.

To ensure the quality of the survey, the provincial and local Center for Disease Control and Prevention created quality control networks to monitor survey efficacy for each selected site. The responsibilities and roles of all staff participating in the survey were defined. A protocol for quality control was developed and implemented. All investigators were trained by qualified trainers before participating in the field survey team. Quality control of the surveyed data was ensured by national and provincial quality control teams. Team leaders reviewed the completed questionnaires before submission for data entry. The field workers taking anthropometric measurements were appropriately trained and evaluated.

The analytical quality control methods for human milk components were to be monitored and evaluated using the NIST SRM 1849a infant formula and pooled breast milk samples prepared in our laboratory for each analysis.

RESULTS AND DISCUSSIONS

Sample distribution among different sites

The total breast milk samples collected were 6,481, of which colostrum, transitional milk and mature milk samples were 1,859, 1,235 and 3,387 respectively. The distribution of sample amounts from different sites is shown in Table 1.

Distribution of sample volume during different breast stages

At different stage of lactation, the amounts and distribution of collected breast milk from each lactating women are shown in Figure 1. At the stage of colostrum, there were about 50% of milk samples within 10 mL~50 mL and about 70% of colostrum equal to or greater than 10 mL; the sample volumes collected at the transitional and mature milk were mostly over 40 mL (78% and 86%) and there were about 68% and 76% over 50 mL for transitional and mature milk respectively.

Characteristics of study participants

The characteristics of lactating women recruited are shown in Table 2. The mean maternal age was 26.8±4.2 years. The distribution of ethnicities included Han (71.0%), Zang (5.6%), Hui (5.8%), Bai (5.5%), Dai (5.5%), Meng (1.8%), Weiwuer (1.6%) and Zhuang

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### Table 1. Geographical distribution of human milk samples collected at different lactation stage

<table>
<thead>
<tr>
<th>Site</th>
<th>Colostrum</th>
<th>Transitional milk</th>
<th>Mature milk</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gansu</td>
<td>326</td>
<td>217</td>
<td>546</td>
<td>1089</td>
</tr>
<tr>
<td>Heilongjiang</td>
<td>423</td>
<td>276</td>
<td>663</td>
<td>1362</td>
</tr>
<tr>
<td>Beijing</td>
<td>199</td>
<td>146</td>
<td>357</td>
<td>702</td>
</tr>
<tr>
<td>Yunnan</td>
<td>314</td>
<td>212</td>
<td>527</td>
<td>1053</td>
</tr>
<tr>
<td>Guangdong</td>
<td>181</td>
<td>114</td>
<td>341</td>
<td>636</td>
</tr>
<tr>
<td>Shandong</td>
<td>101</td>
<td>68</td>
<td>178</td>
<td>347</td>
</tr>
<tr>
<td>Zhejiang</td>
<td>103</td>
<td>61</td>
<td>142</td>
<td>306</td>
</tr>
<tr>
<td>Inner Mong</td>
<td>12</td>
<td>4</td>
<td>91</td>
<td>107</td>
</tr>
<tr>
<td>Guangxi</td>
<td>0</td>
<td>0</td>
<td>101</td>
<td>101</td>
</tr>
<tr>
<td>Xinjiang</td>
<td>0</td>
<td>0</td>
<td>100</td>
<td>100</td>
</tr>
<tr>
<td>Shanghai</td>
<td>200</td>
<td>137</td>
<td>341</td>
<td>678</td>
</tr>
<tr>
<td>Total</td>
<td>1859</td>
<td>1235</td>
<td>3387</td>
<td>6481</td>
</tr>
</tbody>
</table>

---

![Figure 1. Distribution of human milk volume collected for lactating women](image-url)
About 46.9% of mothers completed 9 years or more education. Lactating women were peasants or farmers (30.0%), house worker (24.1%), professional and technical staffs (11.3%) or service staff (10.4%). There were a relatively high percentage of Cesarean sections (45%). The preterm delivery rate was 5.8%. The mean birth weight of these infants was 3314±459 g and the mean age for these infants was 64.4±92.3 days.

Table 2. Characteristics of study participants

<table>
<thead>
<tr>
<th>Variables</th>
<th>Sample size (n)</th>
<th>Mean±SD or %</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (year)</td>
<td>6463</td>
<td>26.8±4.2†</td>
</tr>
<tr>
<td>Nationality</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Han</td>
<td>4573</td>
<td>71.0</td>
</tr>
<tr>
<td>Zang</td>
<td>359</td>
<td>5.58</td>
</tr>
<tr>
<td>Hui</td>
<td>372</td>
<td>5.78</td>
</tr>
<tr>
<td>Bai</td>
<td>353</td>
<td>5.48</td>
</tr>
<tr>
<td>Dai</td>
<td>351</td>
<td>5.45</td>
</tr>
<tr>
<td>Meng</td>
<td>118</td>
<td>1.83</td>
</tr>
<tr>
<td>Weiwuer</td>
<td>100</td>
<td>1.55</td>
</tr>
<tr>
<td>Zhuang</td>
<td>107</td>
<td>1.66</td>
</tr>
<tr>
<td>Education (%)†</td>
<td>3040 (6480)</td>
<td>46.9</td>
</tr>
<tr>
<td>Profession</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Head of enterprises</td>
<td>225</td>
<td>3.47</td>
</tr>
<tr>
<td>Professional &amp; technical staff</td>
<td>730</td>
<td>11.3</td>
</tr>
<tr>
<td>Office worker</td>
<td>519</td>
<td>8.01</td>
</tr>
<tr>
<td>Service staff</td>
<td>672</td>
<td>10.4</td>
</tr>
<tr>
<td>Peasant</td>
<td>1941</td>
<td>30.0</td>
</tr>
<tr>
<td>Worker</td>
<td>60</td>
<td>0.93</td>
</tr>
<tr>
<td>Military staff</td>
<td>5</td>
<td>0.08</td>
</tr>
<tr>
<td>Houseworker</td>
<td>1557</td>
<td>24.0</td>
</tr>
<tr>
<td>Others</td>
<td>771</td>
<td>11.9</td>
</tr>
<tr>
<td>Pre-pregnancy health examination</td>
<td>5876 (6159)</td>
<td>95.4</td>
</tr>
<tr>
<td>Delivery at the county hospital or more larger hospital</td>
<td>6195 (6474)</td>
<td>95.7</td>
</tr>
<tr>
<td>Cesarean</td>
<td>2910 (6467)</td>
<td>44.9</td>
</tr>
<tr>
<td>Preterm neonates</td>
<td>374 (6476)</td>
<td>5.8</td>
</tr>
<tr>
<td>Lactating women BMI</td>
<td>6451</td>
<td>23.1±3.4‡</td>
</tr>
<tr>
<td>Age for infant (day)</td>
<td>6482</td>
<td>64.4±92.3‡</td>
</tr>
<tr>
<td>Birth weight (g)</td>
<td>6330</td>
<td>3314±459‡</td>
</tr>
</tbody>
</table>

†Finishing equal to or more than 9 year compulsory education.
‡The results were expressed as mean±SD.

Figure 2. Diagram for different indicator analysis sampling from total sample pool of human milk

(1.7%). About 46.9% of mothers completed 9 years or more education. Lactating women were peasants or farmers (30.0%), house worker (24.1%), professional and technical staffs (11.3%) or service staff (10.4%). There were a relatively high percentage of Cesarean sections (45%). The preterm delivery rate was 5.8%. The mean birth weight of these infants was 3314±459 g and the mean age for these infants was 64.4±92.3 days. **Sampling diagram from total samples of human milk for different analysis**

We collected 6,481 human milk samples from 11 Provinces/Municipalities or Autonomous regions. However, we cannot analyze all indicators for whole samples due to limited sample volumes and the expense of analytical costs. The plan for further sampling analysis is shown in Figure 2.
Human milk macronutrient analysis
Macronutrients (protein, fat and lactose), total solid and energy have been analyzed by using human milk analyzer (MIRIS-Human Milk Analyzer, Sweden) in the field sites and we have finished the analysis of more than 4,500 human milk samples. The MIRIS Human Milk Analyzer was based on an IR-technology (Infrared transmission spectroscopy) and the standard reference materials were provided by the machine producer. We have finished the comparison of the accuracy and precision of fresh milk samples analyzed by using HMA in the field with frozen samples by classical chemical methods in the our laboratory which showed that HMA might be used to analyze macronutrients in fresh human milk with acceptable accuracy and precision after recalibrating fat and protein level in HMA in field setting. Protein, fat, lactose, and total solid contents in human milk measured by MIRIS Human milk analyzer (HMA) were compared with the classical methods for protein (micro- Kjeldahl method (n=99), Roses-Crottileb extraction method for total fat (n=89), high-performance anion exchange chromatography for lactose (n=100), and direct drying method at temperature 105°C for total solid (n=37) respectively. Compared with the classical chemical methods, the mean protein content was significantly lower and the mean fat level was significantly higher when measured by the HMA method (1.04 g/100g vs 1.22 g/100g and 3.57 g/100g vs 3.13 g/100g, respectively, p<0.001); there were no significant differences for lactose and total solid between the two methods (6.58 g/100g vs 6.52 g/100g, p>0.05 for lactose, and 12.1 g/100g vs 12.0 g/100g, p>0.05 for total solid). The comparison of the methodology have been published elsewhere.25

Analysis of mineral contents in human milk
Mineral analysis method has been developed and finalized by using ICP-MS with microwave digestion. Minerals included Na, Mg, K, Ca, Al, Fe, Zn, Mn, Cu, P, As, Se, Mo, V, Cd, Cr, Co, Ni, Ga, Ag, Sr, Cs, Ba and Pd. The detection limit was from 0.01 µg/kg for As, Mg, V, Si, Ag and Cd to 2.40 µg/kg for phosphorus (P).24 The detection limits for other minerals are within the labeling range. This method had higher accuracy and good repeatability. Total of more than 2,000 human milk samples have been randomly selected for mineral analysis. The levels of minerals in human milk were related to several factors including region, ethnicity, education level, waist to hip ratio of lactating women postpartum and the dietary intakes.

Free B-vitamin analysis in human milk
A high-throughput method for simultaneously analyzing the free forms of B-vitamins in human milk was further developed and validated using ultra-performance liquid chromatography tandem mass spectrometry (UPLC-MS/MS), which included thiamin, riboflavin, pyridoxal, nicotinamide, flavin adenine dinucleotide (FAD), biotin, nicotinic acid, pyridoxine, pyridoxamine and pantothenic acid. The analysis of 1,800 human milk samples has been finished and the findings indicate that the concentrations of free B-vitamins in human milk in china were gradually increased and the contents in colostrum generally much lower than that in transitional milk and mature milk. Further studies will focus on their roles and significance of free B-vitamins in colostrum in neonatal nutrition. The methodology for B-vitamins in human milk at different lactation stages and in various areas of China have been published elsewhere.26

The plan for further analysis
The analyses of human milk in progress include 1,400 samples for fatty acids (C4-C24) and amino acids, 1,200 samples for fatty-soluble vitamins (vitamin A and E) and carotenoids including β-carotene, α-carotene, lutein and zeaxanthin, and about 600 samples for other components including low molecular weight proteins, choline, inositol and taurine. We reserve more than 3,000 human milk samples at -80°C for further analysis of oligosaccharides, oligonucleotides and nucleotides, immunoglobulins and novel active components by region and ethnicity. The program also provides for the assessment of breast milk contaminants and related influencing factors.

Further analyses of 6,481 samples and the survey data will help understand and evaluate the impact of breast on infant growth and development, and establish a human milk composition database by region and ethnicity which could be used for long-term monitoring of its links to lifelong health.

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AUTHOR DISCLOSURES
The authors declare that they have no competing interests.

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Original Article

An on-line database for human milk composition in China

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中国人母乳成分在线数据库

背景和目的：了解母乳成分对确定婴儿和乳母的营养素推荐摄入量（RNIs）至关重要。然而，我国仍然缺少全国性的母乳成分。方法和研究设计：采用横断面调查方法，采集了 11 个省市自治区的母乳样品，分析其营养成分。通过问卷调查、体格检查和生化指标评价乳母及其婴儿的营养与健康状况。结果：总共采集了 6,481 份母乳样品，包括初乳（1,859）、过渡乳（1,235）和成熟乳（3,387）。使用母乳成分分析仪测定了超过 4,500 份母乳样品的蛋白质、脂肪、乳糖、总固体和能量；随机选择 2,000 余份样品测定了 24 种矿物质含量；测定了 1,800 份样品中游离 B 族维生素，包括硫胺素、核黄素、吡哆醇、吡哆胺、吡哆醛、尼克酰胺、尼克酸、黄素腺嘌呤二核苷酸（FAD）、生物素和泛酸；分析了约 800 份母乳样品的氨基酸和蛋白质及不同蛋白质组份（α-乳白蛋白、β-酪蛋白和乳铁蛋白）的含量。此外，还测定了 1,200~2,000 例提供乳汁的乳母血清视黄醇和类胡萝卜素组份、25(OH)D、维生素 B-12、叶酸、铁蛋白和生化指标。正在进行的工作：分析 800~1,200 份母乳的脂肪酸（C4-C24）、脂溶性维生素和类胡萝卜素组份的含量。结论：建立区域性母乳成分数据库的工作处在进展阶段，最终努力建成在线可利用的母乳成分数据库。

关键词：母乳、成分、数据库、乳母、中国